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Scintigraphic Evaluation of Patients with Malignant Tumor of the Head and Neck by Thallium-201-chloride (Tl-201) Scintigraphy

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Abstract: The purpose of this study was to evaluate the usefulness of thallium-201-chloride (Tl-201) scintigraphy for the examination of patients with tumors of the head and neck, and to estimate the correlation of the expression of Na⁺/K⁺-ATPase with Tl-201 scintigraphy. Tl-201 scintigraphy was performed in 61 patients with squamous cell carcinoma and 10 patients with benign tumors of the salivary gland. The tumor retention index was obtained from the early and delayed dynamic Tl-201 scintigraphies. The expression of Na⁺/K⁺-ATPase on the cell membrane was evaluated immunohistochemically. Evaluation of correlations between the histopathological tissue differentiation of tumors, the tumor retention index of Tl-201 scintigraphy and the expression of Na⁺/K⁺-ATPase was performed. The tumor retention index of Tl-201 scintigraphy correlated well with the histopathological tissue differentiation of tumors and also showed a good correlation with the expression of Na⁺/K⁺-ATPase. In addition, the expression of Na⁺/K⁺-ATPase demonstrated a close correlation with the histopathological tissue differentiation of malignant tumors. The tumor retention index could be used for the differentiation of malignant tumors from benign tumors and the expression of Na⁺/K⁺-ATPase was estimated as one of the most important factors for Tl-201 accumulation in malignant tumors.

Key words: Thallium-201-chloride, Na⁺/K⁺-ATPase, Tumor retention index, Scintigraphy

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Introduction

Thallium-201-chloride (Tl-201) was first used to evaluate the viability of the myocardium, and has since been introduced to examine tumor lesions in view of its good accumulation in malignant tumors¹⁻⁶. Immediately after its approval in 1994, this scintigraphic agent

was rapidly introduced for the examination of patients with malignant tumors of the head and neck. We have used this agent since 1996 and have examined about 300 cases by Tl-201 scintigraphy. In our previous report⁷, we concluded that the Tl-201 accumulation in malignant tumors was evident in the early phase after the intravenous injection of Tl-201 and the accumulation remained steady in the late phase (a couple of hours after the injection). Furthermore, the accumulation of the late phase sometimes increased rather than that of the early phase. On the other hand, the Tl-201 accumulation of the late phase in benign tumors usually decreased, although the accumulation in the early phase was as distinct as that in malignant tumors. This difference between malignant tumors and benign tumors depended on the accumulation mechanism of Tl-201^{4,5}. Furthermore, this accumulation mechanism was presumed to relate closely with the expression of Na^+/K^+ -ATPase on the cell membrane because of Tl^+ -ion's physical characteristics similar to K^+ -ion⁶. As for Na^+/K^+ -ATPase, the expression of Na^+/K^+ -ATPase was strongly observed in high-grade malignant tumors more than in low-grade ones. Na^+/K^+ -ATPase produces transmembrane concentration gradients for Na^+ -ion and K^+ -ion through the hydrolysis of one molecule of $\text{ATP}^{2,4,5,8}$. However, the mechanism of Tl-201 uptake by Na^+/K^+ -ATPase is not completely understood⁹, and there have been few reports concerning the relationship between the accumulation of Tl-201 and the expression of Na^+/K^+ -ATPase in malignant tumors of the head and neck.

In the present report, we evaluated the relationship between the accumulation of Tl-201 scintigraphy and the expression of Na^+/K^+ -ATPase to clarify the accumulation mechanism of Tl-201 in malignant tumors of the head and neck.

Materials and Methods

1. Patients

Sixty-one patients of 25 females and 36 males with squamous cell carcinoma in the maxilla (3 females and 8 males), the mandible (6 females and 9 males), the tongue (9 females and 12 males), the oral floor (3 females and 5 males) and the buccal mucosa (4 females and 2 males) were used in the present study. Moreover, 10 patients with benign salivary gland tumor of

Table 1 Patient for Tl-201 scintigraphy and Na^+/K^+ -ATPase expression

Diagnosis	female	male
Squamous cell carcinoma	25	36
Benign tumor of salivary gland	4	6

pleomorphic adenoma (3 females and 4 males) and Warthin's tumor (1 female and 2 males) were also included in this study. These 71 patients were examined because of clinically suspected malignant lesions. Their ages ranged from 35 to 88 years old. Patient distribution according to diagnosis and gender is shown in Table 1. They were examined by Tl-201 scintigraphy and the samples of patients with malignant tumor obtained from excised tumor tissue were used for the immunohistochemistry of the expression of Na^+/K^+ -ATPase. The histopathological diagnosis of tissue differentiation was performed by pathologists in the clinical laboratory. We used patients with salivary gland tumors as a benign control of the scintigraphy. These salivary gland tumors have a different cell nature from squamous cell carcinoma. However, it is not inadequate to compare the scintigraphic results between the salivary gland tumors and squamous cell tumors, because the grade of accumulation of Tl-201 chiefly depends on the cell activity, for example, the growth speed of tumors.

2. Scintigraphy

Tl-201 scintigraphy was performed by using the intravenous administration of 74 MBq per 2 mL of Tl-201. An early dynamic scan (for 5 minutes immediately after injection) and delayed dynamic scan (at 2.5 hours after injection) were carried out by using a Gamma View scintillation camera (Hitachi, Ltd., Tokyo, Japan) with a low-energy ultra-high-resolution parallel hole collimator. Two-second scans were obtained continuously for up to 5 minutes. A single 2-second scan constituted a frame data. Two regions of interest (ROI) on each frame, covering both the tumor area (Fig. 1, A) and the contralaterally symmetrical region (control region, Fig. 1, B), were used to evaluate the uptake of Tl-201 in tumor (time activity curve, Fig. 2). The early and delayed retention indexes of the tumor were calculated from the results of each dynamic scan. The early retention index was the count of tumor divided by the count of control in the early dynamic scan (from 4 to 5 min-

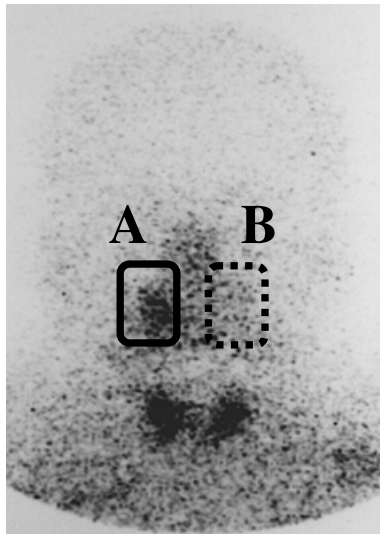


Fig. 1 Two regions of interest (ROI) on a 2-second frame image covering the tumor area (A) and the contralaterally symmetrical region (B, control region).

utes after injection). The delayed retention index was the count of tumor divided by the count of control in the delayed dynamic scan (for 2 minutes at 2.5 hours after injection). Then, the ratio of the delayed retention index to the early retention index was calculated as the tumor retention index. These retention indexes were obtained with reference to reports in the literature.^{7,10}

3. Immunohistochemistry

Immunohistochemical staining was performed with histopathological samples taken from patients. Briefly, 10% formalin-fixed paraffin embedded sections ($5\ \mu\text{m}$) of the resected tumor were mounted on adherent glass slides (Matsunami Glass Ind. Ltd., Japan). Sections were deparaffinized in Hemo-clear (S & S Company of Georgia Inc., U.S.A.) and rehydrated in graded ethanols. Slides were treated with 0.1-mol/L sodium citrate buffer (pH 6), heated in an autoclave at 121°C for 10 minutes for the antigen retrieval, and then treated with 2% hydrogen peroxide for 10 minutes to block endogenous peroxidase activity. Sections were incubated with the primary monoclonal antibody (anti-mouse Mab Na^+/K^+ -ATPase α , M7-PB-E9, Affinity Bio. Inc., U.S.A.) in a moist chamber at room temperature overnight, and then incubated with secondary monoclonal antibody (Biotinized anti-IgG Ab, Affinity Bio. Inc., U.S.A.) for 30 minutes at room temperature. After incubation, the sections were washed with TBS (Tris-buf-

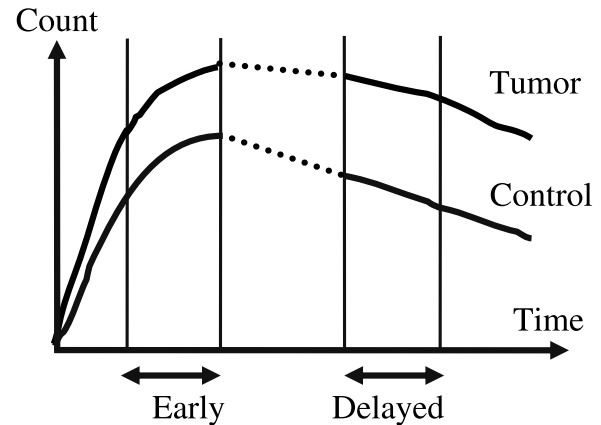


Fig. 2 Time activity curve. The early and delayed retention indexes of the tumor were calculated from the radioactive count shown by the two curves. The early retention index was the count of tumor / the count of control in the early dynamic curves (from 4 to 5 minutes after injection). The delayed retention index was the count of tumor / the count of control in the delayed dynamic curves (for 2 minutes at 2.5 hours after injection). The ratio of the delayed retention index to the early retention index was calculated as the tumor retention index.

fer Saline, DAKO, U.S.A.), reacted with ABC (Avidin-Biotinylated-Peroxidase Complex, Vector Lab. Inc., U.S.A.), and stained with DAB (Diaminobenzidine, Dojindo Lab., WAKO, Japan)^{11,12}. The sections were lightly counterstained with hematoxylin and cover-slipped. The negative control consisted of omission of the primary monoclonal antibody using a case of squamous cell carcinoma, and the positive control was obtained using a section of the kidney because Na^+/K^+ -ATPase distributes physiologically and abundantly on their cell membranes¹³. Na^+/K^+ -ATPase expression of the immunohistochemistry was graded visually on the photomicroscopy into score-0 (stained under 5%), score-1 (from 5 to 49%), or score-2 (over 50%)^{8,12} with reference to the negative (Fig. 3, A) and positive (Fig. 3, B) controls.

4. Statistical evaluation

A chi-square test was performed for the statistical evaluation of the correlation between the histopathological tissue, the tumor retention index of Tl-201 scintigraphy and the expression of Na^+/K^+ -ATPase.

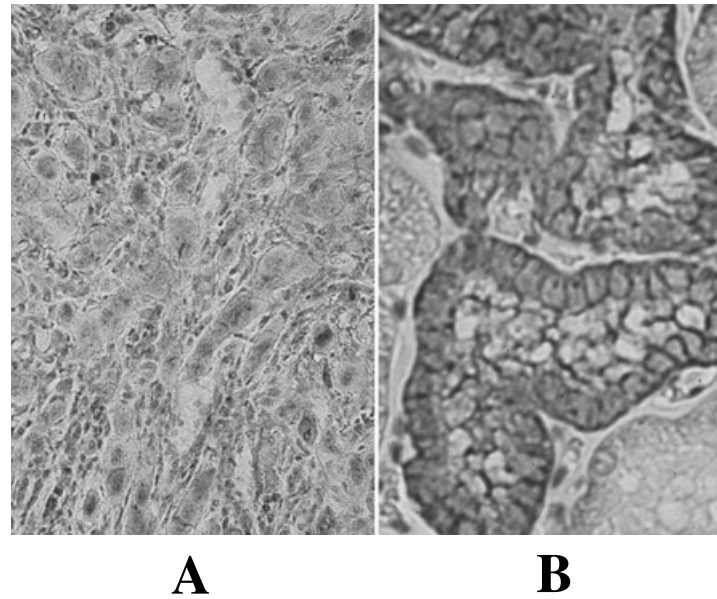


Fig. 3 Immunohistochemical staining of Na^+/K^+ -ATPase. Negative control (A) consists of omission of the primary monoclonal antibody and the positive control (B) is obtained using the section of the kidney because Na^+/K^+ -ATPase showed a physical and distinct distribution on its cell membrane.

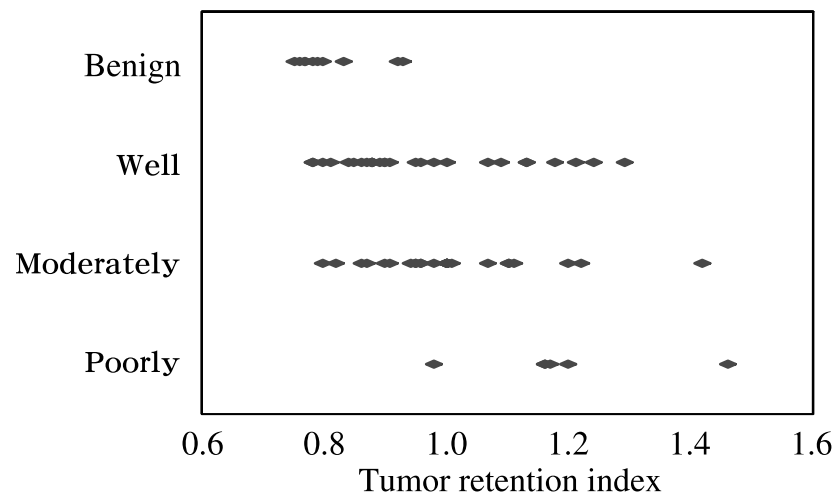


Fig. 4 Distribution of tumor retention indexes of 71 patients with benign and malignant tumors. In patients with benign tumor, the tumor retention indexes ranged from 0.75 to 0.93. In patients with malignant tumor, the tumor retention indexes ranged from 0.78 to 1.29 in the well-differentiated group, from 0.80 to 1.42 in the moderately differentiated group, and from 0.98 to 1.46 in the poorly differentiated group.

Results

1. Distribution of tumor retention index of Tl-201 scintigraphy

Tumor retention indexes of 71 patients with benign

and malignant tumors were distributed as shown in Fig. 4. In patients with benign tumor, the tumor retention indexes ranged from 0.75 to 0.93 with the average \pm SD of 0.810 ± 0.0613 . In patients with malignant tumor, the tumor retention indexes ranged from 0.78 to

Table 2 Tumor retention index and tissue differentiation

		Tumor retention index*, **		
		Decreased < 0.9	Unchanged 0.9-1.1	Increased 1.1 <
Benign tumor		8 (80%)	2 (20%)	0 (0%)
Malignant tumor (Tissue differentiation)	Well	15 (54%)	7 (25%)	6 (21%)
	Moderately	5 (19%)	16 (60%)	6 (22%)
	Poorly	0 (0%)	1 (17%)	5 (83%)

* with benign tumor: chi-square = 28.53 > 22.458 (p=0.001)

** without benign tumor: chi-square = 19.66 > 18.468 (p=0.001)

Table 3 Tissue differentiation and Na⁺/K⁺-ATPase expression

		Na ⁺ /K ⁺ -ATPase expression*		
		Score-0	Score-1	Score-2
Tissue differentiation	Well	10 (36%)	13 (46%)	5 (18%)
	Moderately	0 (0%)	15 (56%)	12 (44%)
	Poorly	1 (16.6%)	1 (16.6%)	4 (66.6%)

* chi-square = 16.17 > 14.86 (p=0.005)

Table 4 Tumor retention index and Na⁺/K⁺-ATPase expression

		Na ⁺ /K ⁺ -ATPase expression*		
		Score-0	Score-1	Score-2
Tumor retention index	Decreased	8 (40%)	9 (45%)	3 (15%)
	Unchanged	1 (4%)	14 (58%)	9 (38%)
	Increased	2 (12%)	6 (35%)	9 (53%)

* chi-square = 13.39 > 13.277 (p=0.01)

1.29 in the well-differentiated group with the average \pm SD of 0.962 ± 0.144 , from 0.80 to 1.42 in the moderately differentiated group with the average \pm SD of 1.00 ± 0.127 , and from 0.98 to 1.46 in the poorly differentiated group with the average \pm SD of 1.18 ± 0.140 .

From the results, the tumor retention indexes were classified into three groups with reference to our previous report⁷: decreased (< 0.9), unchanged (0.9-1.1) and increased (1.1 <) in Table 2. Most patients (8/10 patients, 80%) in benign tumors belonged to the decreased group. On the other hand, most patients of the well-, moderately and poorly differentiated groups in malignant tumors were found in the decreased group (15/28 patients, 54%), the unchanged group (16/27 pa-

tients, 60%) and the increased group (5/6 patients, 83%) respectively. Thus, most patients of the poorly differentiated group belonged to the increased group of the tumor retention index, although most patients with benign tumor and most patients of the well-differentiated group were found in the decreased group of the retention index. A statistical analysis with the chi-square test was carried out. The results indicated a significant correlation between the tumor retention index and the histopathological tissue differentiation (p=0.001).

2. Histopathological tissue differentiation of malignant tumor and Na⁺/K⁺-ATPase expression

This evaluation was carried out in patients with ma-

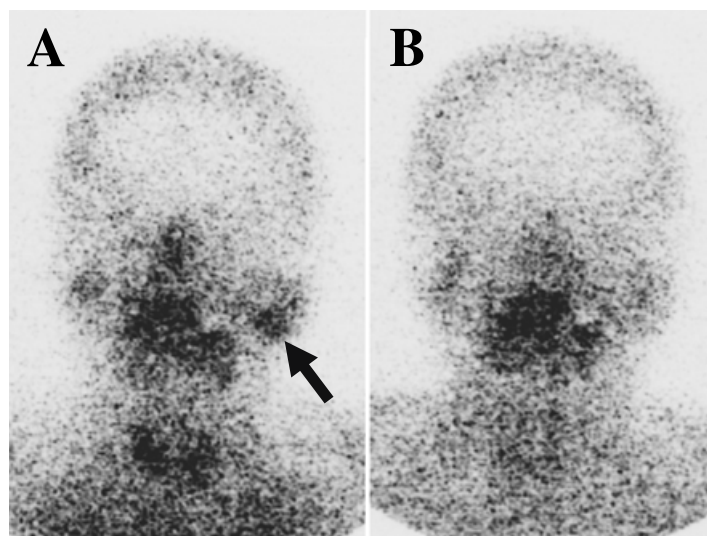


Fig. 5 Case 1: Patient with pleomorphic adenoma in the lower part of the left parotid gland. Early spot image of Tl-201 scintigraphy showed a warm uptake (A, arrow). However, the uptake completely disappeared in the delayed image (B).

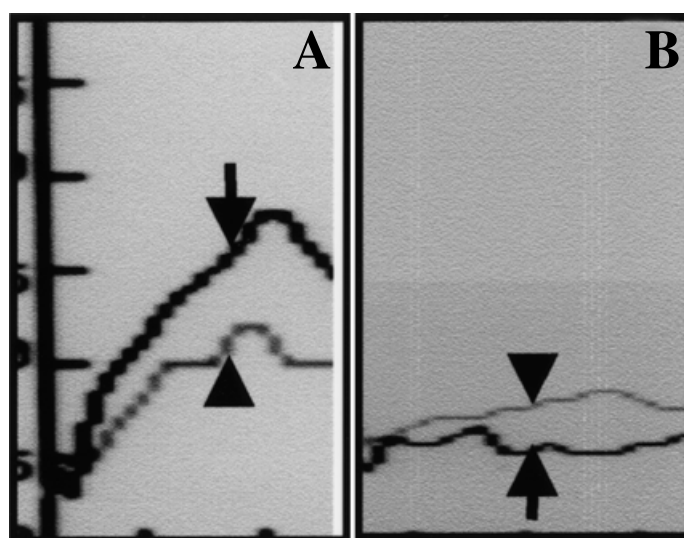


Fig. 6 The time activity curves of case 1 demonstrated the change of difference of radioactive count between the tumor and the control from the early phase (A; tumor, arrow; control, arrowhead) to the delayed phase (B).

lignant tumor (Table 3). Scores-0, 1 and 2 were evaluated with reference to the controls on the photomicroscopy. In the well- or moderately differentiated group (55 patients), most patients (38/55 patients, 69%) had a tendency to show score-0 or score-1. However, most patients of the poorly differentiated group belonged to the score-2 group (4/6 patients, 66.6%). The frequency of score-2 distinctly increased with higher grade of histopathological tissue differentiation. There was a statisti-

cal correlation between the histopathological tissue differentiation of malignant tumor and Na^+/K^+ -ATPase expression ($p = 0.005$).

3. Tumor retention index of Tl-201 scintigraphy and Na^+/K^+ -ATPase expression in patients with malignant tumor

This evaluation was performed in patients with malignant tumor (Table 4). In the decreased or un-

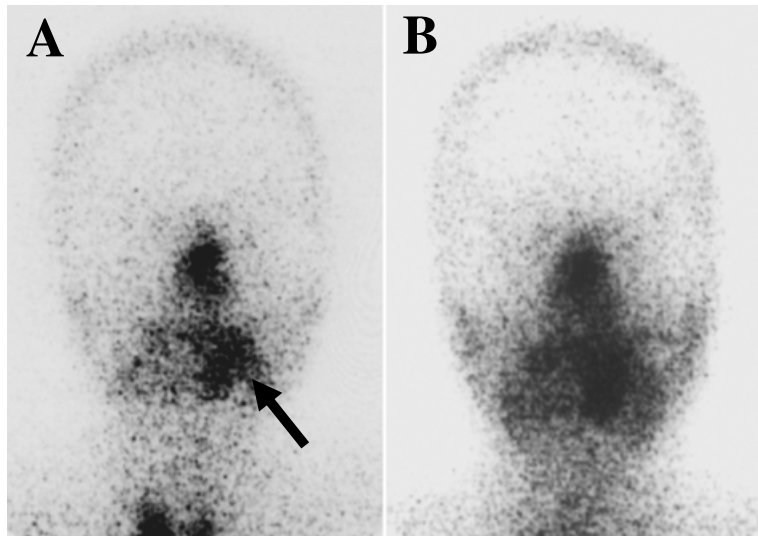


Fig. 7 Case 2: Patient with squamous cell carcinoma of the left oral floor. Tl-201 scintigraphy showed a marked accumulation of the tumor in the early spot image (A, arrow). The accumulation in the delayed spot image (B) did not show any decrease or was rather more distinct than that in the early spot image (A).

changed tumor retention index group (44 patients), most patients were included in the score-0 or score-1 group (32/44 patients, 73%). On the other hand, most patients of the increased tumor retention index group showed score-2 (9/17 patients, 53%). Thus, the frequency of score-2 increased with larger tumor retention index. There was a statistical correlation between the tumor retention index of Tl-201 scintigraphy and Na^+/K^+ -ATPase expression ($p = 0.01$).

4. Case presentation

Case 1: Patient with benign salivary gland tumor of pleomorphic adenoma in the lower part of the left parotid gland. Early spot image of Tl-201 scintigraphy showed a warm uptake (Fig. 5, A, arrow). However, the uptake completely disappeared in the delayed image (Fig. 5, B). The time activity curves in Fig. 6 demonstrate the change of difference of radioactive count between the tumor and the control from the early phase (Fig. 6, A, tumor; arrow, control; arrowhead) to the delayed phase (Fig. 6, B).

Case 2: Patient with squamous cell carcinoma of the left oral floor. Tl-201 scintigraphy showed a marked accumulation of the tumor in the early spot image (Fig. 7, A, arrow). The accumulation in the delayed spot image (Fig. 7, B) did not show any decrease or was rather distinct compared to that in the early spot image. The

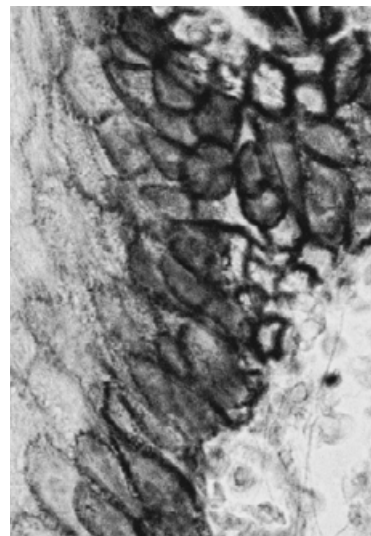


Fig. 8 The expression of Na^+/K^+ -ATPase of case 2 was clear on the cell membrane and estimated as score-2.

tumor retention index of Tl-201 scintigraphy was 1.17 (belongs to the increased tumor retention index group). The expression of Na^+/K^+ -ATPase was clear on the cell membrane and estimated as score-2 (Fig. 8).

Discussion

We showed the usefulness of Tl-201 scintigraphy in our previous report⁷. We also indicated that the tumor retention index of Tl-201 scintigraphy provided supple-

mentary but important information to predict the grade of histopathological tissue differentiation. Some previous reports¹⁴⁻¹⁶ demonstrated almost the same results as ours that the tumor retention index of Tl-201 scintigraphy was useful to differentiate benign tumors from malignant tumors, and it was closely related to the grade of histopathological tissue differentiation. Saijo and co-workers¹⁷ reported that the prognosis of patients with adenocarcinoma was better among patients with well-differentiated tumor than those with poorly differentiated tumor. Therefore, this tumor retention index of Tl-201 scintigraphy could be a supplementary factor to predict the prognosis of patients. In the present report, we evaluated the usefulness of the tumor retention index of Tl-201 scintigraphy with 71 patients including 61 patients with malignant tumor and 10 patients with benign tumor. The tumor retention index of Tl-201 scintigraphy of the present study showed a good correlation with the histopathological tissue differentiation of tumors. The tumor retention index of Tl-201 scintigraphy tended to be larger in malignant tumors than in benign lesions, and furthermore, the tumor retention index became large with higher grade of histopathological differentiation^{7,14-16}. Thus, we think that the tumor retention index is one of the important methods to predict the prognosis of patients with malignant tumors of the head and neck. As for the accumulation mechanism of Tl-201⁴⁻⁶, there are two steps: in the first step the intravenously injected Tl-201 accumulates around tumor cells from the injected site, and in the second step Tl-201 goes through the cell membrane^{18,19}. The injected Tl-201 flows and reaches the tumor cells through the tumor vascular system. Tl-201 accumulation of the first step mainly depends on the volume of blood flow and blood pool⁷. Because the volume of blood flow is generally rich in malignant tumors²⁰ and the tumor vascular system has many fenestrae leading to high permeability, Tl-201 easily moves to tumor cells. Also due to the decrease in blood flow in malignant tumors, Tl-201 accumulates in tumor cells²¹, as a result of which, a lot of Tl-201 accumulates around tumor cells. There are some factors concerning Tl-201 transportation inside tumor cells. Tl-201 has physical characteristics similar to potassium because of its ion-radius being similar to potassium⁷. Tl-201 is a potassium analog and has five times the affinity to cell as potassium⁸. As for the role of Na^+/K^+ -ATPase in the transportation of

Tl-201 into tumor cells, Na^+/K^+ -ATPase is distributed widely in malignant tumor cells²²⁻²⁵ and probably plays one of the most important roles^{6,11-13}. In the present study, Na^+/K^+ -ATPase expression in patients with squamous cell carcinoma was classified into three grades of scores-0, 1, and 2 immunohistochemically. Most patients of the well-differentiated group showed score-0 (36%) and score-1 (46%). Score-2 was shown in only 18% of patients. In contrast, patients of the poorly differentiated group showed score-2 in 4/6 patients (66.6%) and score-0 in only 1/6 patients (16.6%). Thus, Na^+/K^+ -ATPase expression closely was related to the grade of histopathological tissue differentiation of tumors. Furthermore, patients revealed almost the same results between the tumor retention index and the expression of Na^+/K^+ -ATPase²³. Patients showed an obvious difference among the three tumor retention index groups concerning the expression of Na^+/K^+ -ATPase. Score-2 was observed in 15%, 38% and 53% of patients of the decreased, unchanged and increased tumor retention index groups. Based on these results, there was no doubt that the expression of Na^+/K^+ -ATPase was higher in the high-grade malignant tumors than the low-grade malignant tumors and that Na^+/K^+ -ATPase played one of the most important roles in Tl-201 transportation into tumor cells. This speculation was supported by an *in-vitro* experiment that the active transport of Tl-201 into malignant tumor cells was regulated by Na^+/K^+ -ATPase^{20,21}. However, the expression of Na^+/K^+ -ATPase showed a difference between the histopathological tissue differentiation and the tumor retention index. It had a higher correlation with the histopathological tissue differentiation than the tumor retention index of Tl-201 scintigraphy. This difference probably arises because the tumor retention index was affected remarkably by the tumor vascular system, for example, the volume of blood flow, the blood pool, the amount of tumor vessels, and the vascular permeability. We reported in our previous report⁷ that the tumor retention index revealed a good correlation with the tumor vascular system. Thus, the tumor vascular system also plays an important role in the Tl-201 scintigraphy the same as the expression of Na^+/K^+ -ATPase.

In conclusion, the transportation of Tl-201 into tumor cells would depend on the expression of Na^+/K^+ -ATPase. The tumor retention index of Tl-201 scintigraphy can thus be used as an adjunct in predicting the

histopathological tissue differentiation of tumors of the head and neck.

References

- Hisada K., Tonami N., Miyamae T., Hiraki Y., Yamazaki T., Maeda T., et al.: Clinical evaluation of tumor imaging with 201-Tl chloride. *Radiology* 129 : 497-500, 1978.
- Brismar T., Collins P., and Kesselberg M.: Thallium-201 uptake relates to membrane potential and potassium permeability in human glioma cells. *Brain Res* 500 : 30-36, 1989.
- Momose M., Sone S., Maruyama Y., Oguchi K., and Ito A.: Performance of degree of accumulation with 201-Tl-SPECT in 13 cases of lung cancer smaller than 2 cm. *NIPPON ACTA RADIOLOGICA* 60 : 699-701, 2000.
- Carlin R.D., and Jan K.: Mechanism of thallium extraction in pump perfused canine hearts. *J Nucl Med* 26 : 165-169, 1985.
- Mori K., Yamaguchi T., and Maeda M.: Mechanism of 201-Thallium-chloride uptake in tumor cells and its relationship to potassium channels. *Neurological Research* 20 : 19-22, 1998.
- Woo A.L., James P.F., and Lingrel J.B.: Sperm mobility is dependent on a unique isoform of the Na, K-ATPase. *J Biol Chem* 275 : 20693-20699, 2000.
- Sato T., Indo H., Kawabata Y., Iwashita Y., Morita Y., Noikura T., Sugihara K., and Mimura T.: Dynamic scintigraphy with thallium-201 chloride (Tl-201) for the diagnosis of tumors of the head and neck. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 92 : 228-235, 2001.
- Takekawa H., Itoh K., Abe S., Ogura S., Isobe H., Furudate M., and Kawakami Y.: Thallium-201 uptake, histopathological differentiation and Na-K ATPase in lung adenocarcinoma. *J Nucl Med* 37 : 955-958, 1996.
- Lorberboym M., Mandell L.R., Mosesson R.E., Germano I., Lou W., DaCosta M., et al.: The role of thallium-201 uptake and retention in intracranial tumors after radiotherapy. *J Nucl Med* 38 : 223-226, 1997.
- Tonami N., Shuke N., Yokoyama K., Seki H., Takayama T., Kinuya S., et al.: Thallium-201 single photon emission computed tomography in the evaluation of suspected lung cancer. *J Nucl Med* 30 : 997-1004, 1989.
- Abbot A., and Ball W.J.: The epitope for the inhibitory antibody M7-PB-E9 contains Ser-646 and Asp-652 of the sheep Na⁺, K⁺-ATPase α -subunit. *Biochemistry* 32 : 3511-3518, 1993.
- Espineda C., Seligson D.B., Ball W.J., Rao J.Y., Palotie A., Horvath S., et al.: Analysis of the Na, K-ATPase α - and β -subunit expression profiles of bladder cancer using tissue microarrays. *Cancer* 97 : 1859-1868, 2003.
- Abbott A., and Ball W.J.: The inhibitory monoclonal antibody M7-PB-E9 stabilizes E₂ conformational state of Na⁺, K⁺-ATPase. *Biochemistry* 31 : 11236-11243, 1992.
- Ochi H., Sawa H., Fukuda T., Inoue Y., and Nakajima H.: Thallium-201-chloride thyroid scintigraphy to evaluate benign and/or malignant nodules. *Cancer* 50 : 236-240, 1982.
- El-Desouki M.: Thallium-201 thyroid imaging in differentiating benign from malignant thyroid nodules. *Clin Nucl Med* 16 : 425-430, 1991.
- Tonami N., Michigishi T., and Bunko H.: Clinical tumor scanning with 201-Tl-chloride. *Radioisotope* 25 : 829-831, 1976.
- Saijo N., Niitani H., and Tominaga K.: Comparison of survival in nonresected well-differentiated and poorly differentiated adenocarcinoma of the lung. *J Cancer Res Clin Oncol* 97 : 71-79, 1980.
- Ito Y., Muranaka A., Harada T., Matsudo A., Yokobayashi T., and Terashima H.: Experimental study on tumor affinity of 201-Tl-chloride. *Eur J Nucl Med* 3 : 81-86, 1978.
- Sehweil A.M., McKillop J.H., Milroy R., Wilson R., Abdel-Dayem H.M., and Omar Y.T.: Mechanism of 201-Tl uptake in tumors. *Eur J Nucl Med* 15 : 376-379, 1989.
- Kishida T.: Mechanism of thallium-201 accumulation to the thyroid gland: clinical usefulness of the dynamic study in thallium-201 chloride scintigraphy for the differential diagnosis of thyroid nodules. *Kaku Igaku* 124 : 991-1004, 1987 (in Japanese).
- Rogers W., Edlich R.F., Lewis D.V., and Aust J.B.: Tumor blood flow: I. Blood flow in transplantable tumors during growth. *Surg Clin North Am* 47 : 1473-1482, 1967.
- Delpire E., and Gullans S.R.: Cell volume and K⁺ transport during differentiation of mouse erythroleukemia cells. *Am J Physiol* 266 : 513-523, 1994.
- Diomedea L., Bianchi R., Modest E.J., Piovani B., Bubba F., and Salmons M.: Modulation of ATPase activity by cholesterol and synthetic ether lipids in leukemic cells. *Biochem Pharmacol* 43 : 803-807, 1992.
- Numazawa S., Shinoki M.A., Ito H., Yoshida T., and Kuroiwa Y.: Involvement of Na⁺, K⁺-ATPase inhibition in K562 cell differentiation. *J Cell Physiol* 160 : 113-120, 1994.
- Brismar T., and Collins V.P.: Potassium and sodium channels in human malignant glioma cell. *Brain Research* 480 : 259-267, 1989.